



2002 Mosquito-borne Disease Surveillance Program Summary

The Massachusetts Department of Public Health (MDPH) began field surveillance for mosquito-borne viruses in 1957, following outbreaks of eastern equine encephalitis (EEE) in 1955 and 1956, which caused 16 human cases of encephalitis in eastern Massachusetts. EEE virus was first associated with human disease during an earlier outbreak of 35 cases in Massachusetts in 1938. The unpredictable and deadly infection had a case fatality rate of more than 50% during the years after it was first identified, which prompted the United States Public Health Service (USPHS) to fund a study of EEEV at a field station in Taunton, Massachusetts. In 1969, the USPHS discontinued its funding of this field station, and surveillance activities have been continued since by MDPH. The purpose of the Mosquito-borne Disease Surveillance Program is to provide data on the prevalence of mosquito-borne viruses of concern, currently EEE and West Nile viruses, diagnose human illness and provide accurate information for risk assessment and formation of health interventions to reduce the incidence of human disease.

Field surveillance for measuring the prevalence and geographic spread of virus in the environment is done through the collection and testing of mosquitoes, birds, and specimens from ill horses. Surveillance for human disease is done by testing clinical specimens submitted from hospitals for patients with encephalitis, meningitis or other symptoms that may be associated with EEE and WNE.

Avian Surveillance

Certain species of wild birds are the putative enzootic hosts of both EEEV and WNV. The migratory nature of most bird species is one likely means of the introduction and spread of these viruses in the environment. WNV causes severe illness in crows and blue jays, and thus increased deaths of

these species during summer and fall are early indicators of WNV activity in an area. However, these viruses cause infections that are well tolerated in many species, and the avian species that are reservoirs of the viruses during periods of human risk of infection are not known although some species are more suspect based on epidemiological data. Avian surveillance, which consists of dead bird reporting and dead bird testing, is an important component of surveillance.

MDPH operates a toll-free public health information and reporting telephone line (1-866-MASS-WNV), which takes in reports of dead birds during the summer and fall, as well as providing recorded information on WNV infection for the public, and information for human and horse sample submission for Arbovirus testing. Collection of dead bird reports through the information line began May 20, 2002 and allows MDPH to track bird deaths, as well as to test a sample for WNV and EEEV. In 2002, only crows and blue jays were tested since these species are more likely to die from these infections. In order to be suitable for testing, a bird must be reported and received at the laboratory within 24 hours of death. Birds are autopsied to obtain brain tissue that is tested by RT-PCR (Taqman®) using two different primer sets for WNV.

MDPH received over 10,000 dead bird reports during the 2002 surveillance season. More than 850 crows and blue jays were tested from these specimens, and approximately 570 birds tested positive for WNV. In 2001, most of the WNV infected birds were found in the eastern and south-eastern portion of the state with little activity found in western Massachusetts. In 2002, WNV positive bird specimens were submitted from throughout the Commonwealth with infected birds (and human disease) found concentrated in the suburbs north of

Boston, primarily the Malden, Melrose, Medford, and Stoneham areas.

While the number of dead bird reports received in the 2002 season was similar to last year, we tested fewer avian specimens as a result of implementation of a more efficient sampling system, collecting only crows and blue jays for routine WNV testing in 2002. Since crows and blue jays are more susceptible to serious illness and death due to WNV infection, testing of dead birds of these species is a more sensitive means of monitoring virus spread. WNV positive birds were detected for the first time in 37 towns, many of which are located in central and western Massachusetts. A concerted effort was made in 2002 to increase submissions from areas that had previously reported few bird deaths, which may have contributed to the detection of WNV for the first time in some towns.

Mosquito Surveillance

Mosquito collections were done from May 26 until October 4, 2002. Out of a total of 6438 mosquito pools tested on cell culture, (representing 100,513 mosquitoes) 67 pools tested positive for West Nile virus (WNV), and 1 pool tested positive for Eastern Equine Encephalitis virus (EEEV). The

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majority of WNV positive pools consisted of *Culex species* mosquitoes. The positive EEEV isolate was found in a pool of *Culiseta melanura*.

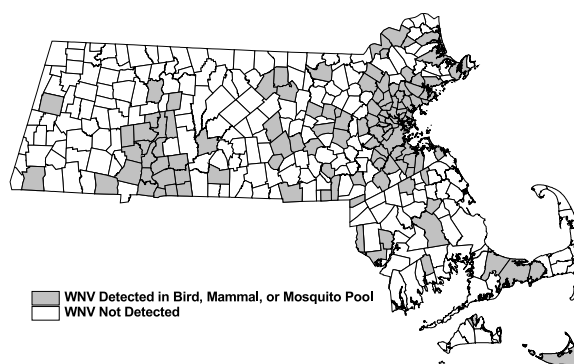
Mosquitoes are collected for testing by staff of MDPH and the regional Mosquito Control Districts. Female mosquitoes are separated from other insects captured in overnight traps and are sorted by species before testing. Species specific information of virus carriage is important for epidemiological analysis and risk assessment of test data. Mosquitoes of the same species are processed in groups ("pools") of up to 50 mosquitoes each. Extracts from pooled specimens are inoculated onto VERO cell cultures. Cell cultures that exhibit cytopathic effect (CPE) are tested by an immunofluorescence method using specific monoclonal antibodies for WNV, EEEV, and Highlands J virus (HJV). Mosquito pools may also be tested by molecular methods such as RT-PCR (Taqman®).

Mosquitoes are collected from seventeen long-term sites that are sampled weekly throughout southeastern Massachusetts (MA) using "miniature" CDC light traps. These trap sites constitute the baseline data for EEEV surveillance and have been sampled since 1957.

Beginning with the detection of WNV in Boston in 2000, field surveillance has added gravid traps and carbon dioxide baited CDC light traps to attract mosquito species that are more likely to carry WNV. Baseline MDPH mosquito collections for WNV have occurred weekly for the past 2 years in Tewksbury, Lowell, Boston, Brookline and Middleboro. In 2002, supplemental mosquito collections were made in ten additional cities and towns in the eastern portion of MA (Woburn, Andover, Saugus, Wakefield, Melrose, Malden, Medford, Revere, Worcester, and Stoneham) and in eighteen cities and towns in the western portion of MA (Ludlow, Springfield, Chicopee, Pittsfield, Westfield, Amherst, Holyoke, Northampton, Agawam, South Hadley, North Adams, Adams, Lenox, Stockbridge, Great Barrington, Belchertown, Hampden and Wilbraham).

WNV activity was detected for the first time in Hampden and Worcester Counties, which had intense positive bird activity as well as positive mosquitoes this season. Much of the positive mosquito activity in Hampden County was concentrated in the Connecticut River Valley region (Chicopee, Springfield and Ludlow), not far from the Connecticut border. Positive mosquito pools in the southeastern portion of MA were found infrequently, with 2 WNV positive pools of mosquitoes from Bristol County in 2002 and none in Plymouth County.

WNV Activity in Massachusetts, 1/1/02 - 11/4/02



The majority of WNV positive mosquito pools came from Norfolk (18 positive pools) and Suffolk (21 positive pools) Counties. Overall, WNV positive mosquito prevalence has risen in five out of fourteen counties in MA, and is higher for the whole state as compared with the past two seasons. This increase in mosquito positivity is in part due to more intensive surveillance efforts, but probably represents a true increase as reflected by the increased incidence of human disease.

Human and Horse Surveillance

There was active collaboration between MDPH and medical providers to ensure that the possibility of human WNV infection was considered among the diagnoses for encephalitis and/or viral meningitis cases. The efforts of the Epidemiology Program in the Communicable Disease Bureau of MDPH and hospital staff assured comprehensive reporting of suspect cases of encephalitis and viral meningitis to the MDPH, and the submission of specimens for evaluation.

For both human and equine serum samples, IgM and IgG enzyme-linked immunosorbent assays (EIA) are used to detect antibody to WNV or EEEV. Plaque reduction neutralization assays are performed to confirm virus-specific antibody responses. CSF is examined for IgM antibody and tested for the presence of virus on cell culture. In some instances, molecular methods may also be used to test for viral RNA.

In 2001, 3 human cases of WNV were diagnosed, 2 in Bristol county and 1 in Middlesex County. In 2002, 24 human cases of WNV were diagnosed in the eastern and central portions of the State. Three of these cases were likely acquired outside of Massachusetts, 2 in Missouri and 1 in Michigan.

The onset dates for the 21 human cases that were likely Massachusetts' cases range from August 13, 2002 to October 1, 2002. Clinically, sixteen of these infections caused meningo-encephalitis and 5 were diagnosed as West Nile fever, a milder form of infection. There were 3 fatalities among the meningo-encephalitis cases, all in patients aged 80 years or older.

Human cases were geographically distributed as far north as Essex County, as far south as Norfolk County and as far west as Worcester County. The majority of cases were detected in Suffolk and Middlesex counties.

In 2001, 45 horses were detected with WNV, the majority in Bristol and Plymouth Counties. The outbreak of WNV in horses prompted many owners to vaccinate their horses with a new WNV equine vaccine. Of the 36 horse specimens submitted to SLU for WNV testing in 2002, one from Ipswich tested positive for WNV. The onset of illness for this horse was October 9, 2002. One horse from Granville, MA tested by the Connecticut Veterinarian Diagnostic Laboratory at the University of Connecticut was also positive for WNV this year. In addition, one llama from Halifax, MA tested positive for WNV early in the season.

Development of a Biomonitoring Plan for Massachusetts

Massachusetts was one of twenty-five states to receive a grant from the Centers for Disease Control and Prevention (CDC) to develop a strategic biomonitoring plan. Biomonitoring is the measurement of the concentration of toxic chemicals or their metabolites in biological specimens, such as blood, serum, urine or saliva to assess human exposure to contaminants in the environment. There are considerable data for toxicants in air, water, soil and food products, but only sparse data for toxicant levels in humans. Data on toxicant levels in environmental and food samples provide an estimate of potential exposure. However, this "external dose" or potential exposure is not a direct measure of the actual exposure that humans receive, as it does not consider confounding factors, such as the rate of transfer of chemicals across membranes and the different routes of exposure, through which chemicals enter the body.

Biomonitoring or the testing of human specimens is a measure of the effective or "internal dose" of environmental contaminants, which provides more accurate data that can aid in the determination of disease causation through epidemiological studies. The Biomonitoring Grant Program was developed by CDC to increase the capacity and capabilities of the state laboratories to monitor biological specimens for environmental contaminants or their metabolites, and provide data and information for public policy and decision-making.

The State Laboratory Institute (SLI) in collaboration with the Department of Public Health's Bureau of Environmental Health Assessment and the Harvard School of Public Health will develop recommendations for a strategic biomonitoring plan for Massachusetts. The goal of the biomonitoring plan is to provide high quality information that is useful for public health research, a basis for public health decision-making and a source of reference data for medical professionals in evaluating patient exposure. During the planning process, SLI has focused its efforts on designing targeted biomonitoring studies and studies geared towards determining the background levels of environmental contaminants or their metabolites in the general population. The targeted studies will concentrate on areas or population segments where there are an increased prevalence of disease or there has been significant exposure of a population to environmental contaminants. A needs assessment will be performed to aid in the selection of the contaminants for the background biomonitoring studies. The needs assessment will take into consideration the prevalence of the contaminants in Massachusetts, the toxicity of the contaminants, and the at-risk populations.

One of the environmental contaminants under consideration for a background biomonitoring study is environmental tobacco smoke. A brief overview of the issues regarding environmental tobacco smoke

and the biomonitoring planning process provides both an idea of the benefits that can be obtained from the biomonitoring program and the challenges that will be encountered in measuring human exposure. Exposure to environmental tobacco smoke, a known carcinogen, increases the risk for lung cancer and other disease. Children are especially susceptible to environmental tobacco smoke, which may be associated with a higher incidence of bronchitis and pneumonia or the exacerbation of asthma in young children. Cotinine is considered the best biomarker for exposure to environmental tobacco smoke because it remains in the body longer than nicotine and is specific to tobacco smoking. However, high levels of environmental tobacco smoke typically results in concentrations of less than 15 parts per trillion in blood, and are difficult to measure accurately. Furthermore, interpretation of test data requires an adequate database of background levels of cotinine in the general population. Once developed, the database can be used for many purposes, such as tracking the outcomes of the Massachusetts Tobacco Control Program and assessing the exposure of at-risk individuals, such as children of smokers. Biomonitoring programs will aid the development of public health policies, public health research and promote the health of the people of Massachusetts. Contact julianne.nassif@state.ma.us

Packaging and Shipping Diagnostic Specimens and Infectious Substances

The Massachusetts State Laboratory Institute (SLI) offers a one-day training course on packaging and shipping of diagnostic specimens and infectious substances. These materials are classified as dangerous goods and federal regulations require specific packaging and labeling of these materials. Improperly packaging infectious substances for shipment can lead to potential liability both civil and criminal. There are fines associated with penalties for both the shipper and the recipient organization. National and International Regulations require all shippers of hazardous/dangerous goods be trained (See DOT 49 CFR 172.700 and

IATA Dangerous Goods Regulations, 2002 Regulation 1.5). Additionally, in order to ship diagnostic specimens and infectious substances by air, International Air Transport Association Regulations require the shipper to satisfactorily pass a written exam.

The SLI developed a course, "Packaging and Shipping Diagnostic Specimens and Infectious Substances," specifically for **employees of clinical laboratories who prepare or offer for transport specimens by ground or air transportation**. At least one individual responsible for packing and shipping laboratory specimens for transport at a

clinical laboratory should attend the training, which is provided by SLI to Massachusetts' hospitals, clinics and laboratories.

The course provides the shipper with the knowledge and references needed to practice proper techniques for packaging, marking, labeling and documenting shipments of diagnostic specimens and infectious substances. Satisfactory completion of the training assures that each facility that transports specimens can do so in a safe and expeditious manner that ensures timely test results. Carriers often return packages containing

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Packaging and Shipping Diagnostic Specimens and Infectious Substances

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these materials to the shipper due to incorrect labeling or packaging of materials. The program integrates lecture, demonstrations, hands-on training, classroom exercises, and a written examination as required by IATA.

Participants upon completion of the course and passing the exam receive certification of training and will be able to use the materials, references and knowledge acquired in the course as an aid in setting up in-house training for employees at their own facilities.

Participants learn how to identify and distinguish between a diagnostic specimen and an infectious substance; classify specimens according to risk groups and identify and

handle select agents; properly package, mark and label packages; and choose the correct packing materials according to the sample being shipped, the mode of transportation, the carrier, and required shipping documents.

In 2002, courses were held at various locations throughout Massachusetts including Springfield, Holyoke, Newton, Woburn, Fall River and Jamaica Plain. Additional courses are being scheduled for 2003. The course will be offered once a month beginning in February. Most courses can accommodate 20 to 30 participants depending upon the location. The course is provided free to

employees of clinical laboratories in Massachusetts and pre-registration is required at least one week prior to the course date.

Course announcements for 2003 will be mailed to clinical laboratories, published in the SLI Newsletter, and posted on our website. If you are interested and wish to be placed on our mailing list for future notification, please contact Garry Greer, State Laboratory Training Coordinator at 617-983-6608, or Phyllis Madigan, Chief of Labs at 617-983-6656 or Phyllis.Madigan@state.ma.us and give your complete name, organization, business mailing address, and phone and fax numbers.

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